

LISTING OF CLAIMS

1-10. (Canceled)

11. (Previously Presented) A method for the enzymatic production of terminally or subterminally hydroxylated fatty acids, which comprises

- a) converting a fatty acid selected from terminally saturated, branched or unbranched fatty acids with 8 to 30 carbon atoms or fatty acid derivative thereof, selected from C₁-C₄ alkyl esters, amides and anhydrides, in the presence of an electron donor system, a cytochrome P450 monooxygenase and oxygen wherein said electron donor system comprises an inorganic, non-electrode bound source of electrons and a mediator which is able to transfer electrons from the source of electrons to the enzyme, wherein said enzyme is a cytochrome P450-containing mono-oxygenase (E.C. 1.14) of the families CYP4, CYP52, CYP102, and wherein the source of electrons is a metal in powder form with a lower standard normal potential than the mediator; and
- b) isolating the hydroxylated product(s).

12. (Previously Presented) A method as claimed in claim 11, wherein the ω -hydroxylatable fatty acid derivative is selected from terminally saturated, branched or unbranched C₁₀-C₃₀fatty acids.

13. (Canceled)

14. (Currently Amended) A method as claimed in claim 11, wherein the cytochrome P450 mono oxygenase has a modification in the amino acid sequence SEQ ID NO:35, which modification consists of a single mutation wherein F87 is replaced by A or V, L188 is replaced by K, V26 is replaced by T, R47 is replaced by F, or V26 is replaced by T is a single mutant selected from the group consisting of F87A, F87V, L188K, V26T, R47F and V26F.

15. (Currently Amended) A method as claimed in claim 11, wherein the cytochrome P450 mono oxygenase has a modification in the amino acid sequence SEQ ID NO:35, which modification consists of a mutation wherein F87 is replaced by A or V, and one further modification wherein L188 is replaced by K, A74 is replaced by G, R47 is replaced by F and V26 is replaced by T ~~is a mutant having in position 87 the mutation F87A or F87V and at least one other of the following mutations: L188K, A74G, R47F and V26T.~~
16. (Previously Presented) A method as claimed in claim 26, wherein the electron donor system is zinc/Co(III) sepulchrate.
17. (Previously Presented) A method as claimed in claim 11, wherein at least stage a) is carried out in the presence of chloride ions.
18. (Previously Presented) A method as claimed in claim 11, wherein at least stage a) is carried out in the presence of a hydrogen peroxide-cleaving enzyme.
- 19-22. (Canceled)
23. (Previously Presented) A method as claimed in claim 11, wherein the mediator has a standard normal potential in the region of less than about -0.4 V.
24. (Previously Presented) A method as claimed in claim 11, wherein the mediator is selected from cobalt(III) sepulchrate, methylviologen, neutral red, riboflavin, ruthenium triacetate, FMN and FAD.
25. (Previously Presented) A method as claimed in claim 11, wherein the source of electrons is metallic zinc.

26. (Previously Presented) A method as claimed in claim 11, selected from the systems:

- Zn/cobalt(III) sepulchrate and
- Zn/neutral red.

27. (Previously Presented) A method as claimed in claim 12, wherein the ω -hydroxylatable fatty acid derivative is selected from terminally saturated, branched or unbranched C₁₂-C₃₀ fatty acid.

28. (Previously Presented) A method as claimed in claim 12, wherein the ω -hydroxylatable fatty acid derivative is selected from terminally saturated, branched or unbranched C₁₂-C₂₅ fatty acid.

29. (Previously Presented) A method as claimed in claim 12, wherein the ω -hydroxylatable fatty acid derivative is selected from terminally saturated, branched or unbranched C₁₂-C₂₀ fatty acid.

30. (Previously Presented) A method as claimed in claim 11, wherein the cytochrome P450 mono oxygenase is a mutant, which is obtained by amino acid substitution in at least one of positions 26, 47, 72, 74, 87, 188 and 354, of the wild-type enzyme (SEQ ID NO: 35).